Nutritional Factors and Susceptibility to Lead Toxicity

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Although the quantities of lead (Pb) to which individuals are exposed vary widely, susceptibility of an individual to the effects of a specific level of exposure is another highly important factor in development of lead toxicity. For example, susceptibility to lead toxicity can be modified by several dietary factors. Low dietary intakes of calcium or iron (20% of recommended levels) substantially increase the toxicity of the same level of lead exposure to rats. In the studies of calcium effect, when calcium was fed to rats at 1/5 of the recommended intake, 12 μg Pb/ml drinking water produced the same degree of toxicity as did 200 μg Pb/ml with a normal calcium diet. The maximal dose for a 10-week period that does not impair heme synthesis or renal function in the rat has been established to be 200 μg Pb/ml drinking water. The role of low calcium diet on increasing susceptibility to lead has been confirmed in several species.

Mechanisms explaining the effect of calcium on lead toxicity may be related to absorption of lead from the gastrointestinal tract or renal tubule or to function of the parathyroid. Preliminary histological investigations on the parathyroids of control and lead-treated rats on normal and low calcium diets show no effect of lead.

Studies are currently underway to evaluate the lead, calcium and iron contents of the diets of children with normal and elevated concentrations of blood lead.

Susceptibility to lead (Pb) toxicity is known to be influenced by a number of physiological and environmental factors (*1*): (1) age; (2) season of the year (body temperature, dehydration, ultraviolet light); (3) calcium, phosphorus and vitamin D; (4) dietary protein; (5) ascorbic acid; (6) nicotinic acid; (7) alcohol; (8) other heavy metals. The types of factors involved in alteration of vulnerability vary widely but include dietary and metabolic effects. The relative importance of these factors under similar experimental or environmental conditions has yet to be defined.

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The effects of low dietary calcium (Ca) or iron (Fe) concentrations on susceptibility to Pb toxicity have recently been investigated (*2,3*). In these studies, rats received 200 μg Pb/ml drinking water for 10 weeks. Previous studies (*4*) have shown that this Pb level is the maximum concentration of Pb which, when given orally for 10 weeks, does not produce significant alteration in hematopoiesis or renal size, histology, or function of rats fed a Purina Laboratory Chow diet. In these experimental studies the low dietary levels of Ca or Fe used were one-fifth of the recommended daily allowances for the growing laboratory rat (*5*). The phosphorous concentrations of

the diets used in the Ca and Fe studies were constant.

The studies showed that a low Ca diet results in increased susceptibility to Pb toxicity, for example, it increased blood Pb concentration of rats exposed to Pb approximately fourfold (Fig. 1). The concentrations of Pb in soft tissues such as kidney and in bone are much higher on low Ca diets (*2*). However, the increase in Pb content is much greater for soft tissue than for bone. The influence of Ca was also demonstrated (*2*) for biological effects indicative of Pb exposure, for example, urinary excretion of δ-aminolevulinic acid (δ-ALA); excretion of δ-ALA when lead is given much higher on a low Ca diet than on a normal Ca diet.

The magnitude of the Ca effect has been studied in rats (*6*). The effects of different doses of Pb on renal Pb content of animals fed low and normal Ca diets are compared in Table 1. Concentraitons of Pb in drinking water needed to produce similar degrees of Pb effect on normal and low Ca diets are compared in Table 2. Other parameters of Pb effect, such as bone and kidney Pb concentrations, are comparable at 12 μg Pb/ml on low Ca diets with 200 μg Pb/ml on normal Ca diets.

The effects of varying concentrations of dietary Ca on susceptibility to Pb toxicity has been confirmed in a number of species. For example, decreasing the Ca concentration of the diet increases susceptibility to Pb toxicity in the dog, horse, and pig (*7–9*).

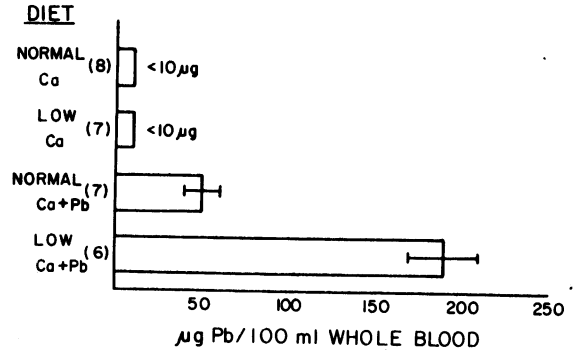

FIGURE 1. Influence of dietary calcium on blood lead concentrations. Data of Mahaffey-Six and Goyer (*2*).

Table 1. Renal Pb content of animals fed normal and low Ca diets with varying concentrations of Pb.

Pb in drinking water, μg/ml	Renal Pb, μg/g wet tissue [a]	
	Normal Ca diet	Low Ca diet
—	1.0±0.1	3.6±0.5
3	1.3±0.1	6.6±1.4
12	1.9±0.1	19.6±2.0
48	5.1±0.4	154 ±51
96	6.9±0.7	629 ±170
200	21.3±3.8	942 ±362
400	20.4±1.9	

[a] Mean ± 1 S. E.

Table 2. The minimal concentration of lead in drinking water of rats fed a low Ca diet which will produce various signs of lead intoxication.

	Minimal toxic dose, μg/ml	
	Low Ca diet	Normal Ca diet
Inclusions	12	200
Urinary δ-ALA	12	200
Kidney weight	3	200

The metabolisms of Ca and Pb are similar in certain respects and have a number of potential sites for interaction. Some of these possible sites are outlined in Figure 2. Because there is an overall increase in Pb re-

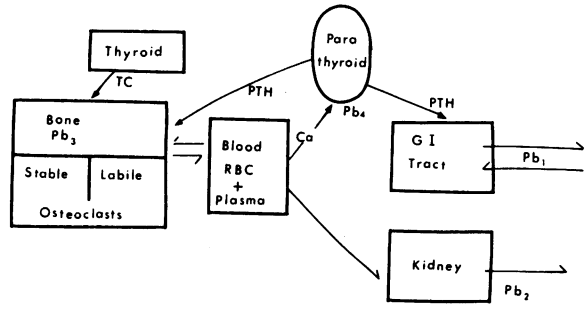

FIGURE 2. Interaction of lead and calcium metabolism. Pb_1, gastrointestinal tract: Pb–Ca competition for absorption; vitamin D increases the absorption of Ca and Pb. Pb_2: increased Ca excretion by kidney due to impaired reabsorption by renal tubule. Pb_3: Pb and Ca storage in bone; effects of parathyroid hormone and thyrocalcitonin. Pb_4: modification of serum Ca level by altered secretion of parathyroid hormone Pb_5: possible modification of PTH by Pb.

tention in animals on low Ca diets, competition between Pb and Ca for active or passive gastrointestinal absorption appears likely; this competition produces an increase in Pb absorption.

Another site of interaction between Pb and Ca is the kidney. Lead is known to damage the proximal renal tubule, producing a Fanconi-type syndrome with impaired reabsorption of glucose, amino acids, and phosphate (*10*). The renal reabsorption of Ca is also likely affected by lead. However, because Ca is reabsorbed throughout the renal tubule, damage to the proximal tubule has less effect on Ca excretion than on excretion of phosphorus, glucose, or amino acids.

Bone is another site of Pb-Ca interaction. A low Ca diet causes an increase in bone Pb content and concentration, but this increase is relatively less than that in Pb content of soft tissue such as kidney. This may be due to the smaller total binding capacity of bone for Pb in animals on a low Ca diet. Hsu et al. (*9*) recently observed lead-containing intranuclear inclusion bodies in the osteoclasts of Pb-treated pigs. These authors suggest that during osteolysis the osteocyte is very active metabolically and responds to Pb intoxication. On continued exposure the osteocyte dies, the area of bone containing Pb becomes necrotic, and the dead bone tissue is resorbed by the osteoclasts. Lead is ingested by the osteoclasts (cells having recognized phagocytic activity), and Pb inclusion bodies develop in these cells. The pathogenesis of these inclusion bodies in the osteoclast is undetermined.

The parathyroid gland represents a fourth side of interaction for Pb and Ca. Lead is known to interfere with the function of some endocrine glands. For example, Sandstead et al. (*11*) reported hypothyroidism in 9 of 16 Pb-poisoned patients. In recent preliminary histological studies on the parathyroids of Pb-treated rats (Goyer and Mahaffey, unpublished data), the parathyroids appeared normal by light microscopy, and the number of cells per visual field was not influenced by Pb. Further histological studies are in progress. Supplementing histological studies by measurement of circulating parathyroid hormone would help to identify any effects of Pb on parathyroid function.

The role of vitamin D in Ca-Pb interactions is not well established. Metabolites of the vitamin act on the gastrointestinal tract, kidney, and bone in regulating Ca metabolism. Similar effects of vitamin D might occur for Pb, but this is only speculative. An early paper by Sobel et al. (*12*) indicated that vitamin D-treated rats had higher tissue Pb concentrations than vitamin D-deficient rats. It is of interest to determine if Pb affects the metabolites of vitamin D such as 25-hydroxycholecalciferol (25-HCC) or 1,25-dihydroxycholecaliferol (1,25-DHCC). Rosen and Roginsky reported (*13*) that the level of 25-HCC in plasma did not differ among groups of children with blood Pb concentrations of <40, 41–60, or 61–126 μg/100 ml. The concentration of 1,25-DHCC, the metabolically active form of vitamin D, was not measured in these children. Lead could affect renal conversion of 25-HCC to 1,25-DHCC. Strontium, which can produce rickets in experimental animals, interfered with the production of 1,25-dihydroxycholecalciferol but did not affect production of 25-hydroxycholecalciferol (*14*). Lead may act in a similar manner.

Iron deficiency, like Ca deficiency, increases susceptibility to Pb toxicity in the rat. This is seen in the effect of Pb on heme synthesis. Table 3 shows hematocrit and urinary δ-ALA values for rats receiving diets containing 5 or 25 ppm Fe and deionized water or solutions of deionized water with 200 μg Pb/ml for a 10-week period. The low Fe diet, although it depleted liver Fe stores, produced little effect on hematocrit. The combination of low Fe intake and Pb exposure acted synergistically to impair heme synthesis. Iron deficiency resulted in increased Pb content of kidney and bone (Table 3). Table 4 shows differential tissue distribution of Pb in rats fed diets deficient in Ca and Fe. Although the amount of Pb in bone is comparable with both types of

Table 3. Hematocrit, urinary δ-aminolevulinic acid (δ-ALA), and renal and femur Pb content of rats fed normal and low Fe diets with and without Pb.

Dietary Group	n	Hematocrit [a]	Urinary δ-ALA, μg/24 hr [a]	Renal Pb, μg/g wet tissue [a]	Femur Pb, μg/g wet tissue [a]
Normal Fe	9	45.7±0.6	20±5	1.0±0.1	5.6±1.4
Low Fe	8	42.6±1.0	22±5	1.9±0.4	10.6±3.0
Normal Fe + Pb [b]	8	44.2±0.9	180±25	14.5±1.6	75.2±13.1
Low Fe + Pb [b]	8	37.8±0.9	355±50	38.7±4.8	225.2±15.2

[a] Mean±1 S. E.
[b] 200 μg Pb/ml drinking water.

Table 4. Comparison of tissue concentrations of lead in rats fed diets deficient in calcium (LCa) and iron (LFe) and nutritionally adequate diets (NCa, NFe).

Diet	Pb, μg/g wet tissue	
	Bone	Kidney
No Pb		
NCa, NFe	2.2±1.0	2.6±1.2
LFe	10.6±3.0	1.9±0.4
LCa	9.7±2.2	4.4±0.6
200 μg Pb/ml H_2O		
NCa, NFe	74±12	22±4.3
LFe	225±15	28.7±4.8
LCa	202±202	691±203

deficiency, kidney Pb is far higher in the Ca-deficient animals.

Figure 3 shows some possible mechanisms by which Fe deficiency may increase Pb uptake and toxicity. Increased Pb absorption from the gastrointestinal tract is a reasonable explanation for the increase in body burden of Pb resulting from Fe-deficient diets. It is known that Fe-deficient diets increase the absorption of certain metals including manganese (*15, 16*), cobalt (*16, 17*), and zinc (*16*) but not cesium, mercury, calcium, or copper (*16*). The relation between Pb and Fe deficiency may be mediated through lowered tissue Fe levels in Fe-deficient animals. Waxman and Rabinovitz (*18*), using incubation of reticulocytes, reported that Pb inhibited hemoglobin synthesis as a specific antagonist of heme synthesis and as a general heavy metal inhibitor of cell metabolism. Ferrous iron protected both these functions against the action of Pb. Lowering of tissue Fe by Fe-deficient diet could increase the inhibitory effects of Pb on heme synthesis. However, until more is known, explanations of Fe-Pb relationships remain speculative.

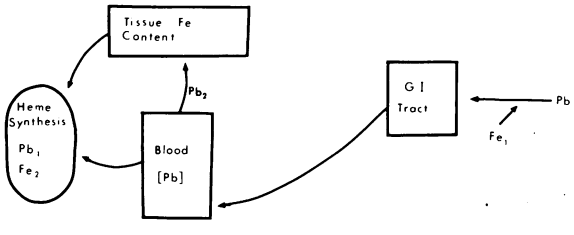

FIGURE 3. Interaction of lead and iron metabolism. Fe: increased Pb absorption in Fe deficiency. Pb_1: inhibition of heme synthesis by Pb. Fe_2: addition of Fe to reticulocytes *in vitro* reduces Pb_1 effect. Pb_2: mild hemolytic anemia in Pb poisoning increases serum Fe and tissue Fe.

The studies described above were carried out in experimental animals. The extent to which these factors affect humans remains unproven. Certainly many young children have prolonged exposure to Pb, and low dietary intakes of Fe or Ca are not rare. The human diet frequently contains lower than recommended amounts of Ca and Fe. Table 5 shows cumulative percentages of children ingesting 1/4, 1/2, and 3/4 of the recommended daily intakes (*19*) of Ca and Fe. The data pertain to children 2–3 years old, studied between 1968 and 1970 in the 10-state nutrition survey (*20*). Certainly low dietary intakes occur more frequently for Fe than Ca; however, based on this and other dietary surveys (*21,22*), Ca intakes of less than 200 mg/day are found in 5 to 15% of children from low income groups. It is also these children who run the greatest risk of exposure to lead.

Table 5. Cumulative percentage distribution of calcium and iron intakes for children 24–36 months of age for low income ratio states.[a]

Calcium		Iron	
Intake, mg/day	Per cent of children	Intake, mg/day	Per cent of children
200	13.7	3.9	24.1
400	28.9	7.9	67.3
600	50.1	11.9	90.9
800[b]	65.9	15.0	98.0

[a] Data are from the 10-State Nutrition Survey, 1968–1970 (*20*).

[b] Recommended daily allowances for children 2 to 3 years-old, 1968 (*19*).

The relationship between Fe and Pb is evident from clinical experience. For example, one way in which children having "asymptomatic" Pb poisoning are identified is that they develop an anemia that does not respond to iron therapy until they are treated for Pb poisoning.

Two reports on Pb content of food were presented at this symposium by Kolbye et al. (*23*) and by Mitchell (*24*). Those data were based on foods prepared in laboratories and analyzed for Pb content. In order to provide an additional assessment of the level of Pb in children's food as consumed in the home, a dietary study was carried out during 1973 at Children's Hospital in Washington, D.C. under a Food and Drug Administration contract. In addition, this study will assess the nutrient content of the total diet of a group of young children and determine if the body burden of Pb as demonstrated by blood Pb concentration correlates with the ingestion of Fe and Ca.

In summary, the role of dietary Ca and Fe in altering the toxicity of Pb is well established in experimental animals. Epidemiological research on the interrelationships of nutritional status and susceptibility to Pb toxicity in children is currently underway. The role of dietary factors is not in the area of treatment of acute Pb poisoning but in the field of preventive medicine. This concept is particularly well illustrated in cases where children after chelation therapy are returned to an environment similar or identical to that in which they developed Pb poisoning. Optimal nutritional status may serve to protect children from such marked adverse effects of excessive Pb exposure.

REFERENCES

1. Goyer, R. A., and Mahaffey, K. R. Suspectibility to lead toxicity. Environ. Health Perspect. No. 1: 73 (1972).
2. Mahaffey-Six, K., and Goyer, R. A. Experimental enhancement of lead toxicity by low dietary calcium. J. Lab. Clin. Med. 76: 933 (1970).
3. Mahaffey-Six, K., and Goyer, R. A. The influence of iron deficiency on tissue content and toxicity of ingested lead in the rat. J. Lab. Clin. Med. 79: 128 (1972).
4. Goyer, R. A., et al. Lead dosage and the role of the intranuclear inclusion body. Arch. Environ. Health. 20: 705 (1970).
5. National Academy of Sciences–National Research Council Publ. 990. Nutrient Requirements of laboratory animals. 1962, p. 71.
6. Mahaffey, K. R., Goyer, R. A. and Haseman, J. K. Dose-response to lead ingestion in rats fed low dietary calcium. J. Lab. Clin. Med. 82: 92 (1973).
7. Calvery. H. O., Laug, E. P., and Morris, H. J. The chronic effects on dogs of feeding diets containing lead acetate, lead arsenate and arsenic trioxide in varying concentrations. J. Pharm. Exper. Therap. 64: 364 (1938).
8. Willoughby, R. A., Thirapatsakun, T., and McSherry, B. J. Influence of rations low in calcium and phosphorus on blood and tissue lead concentrations in the horse. Amer. J. Vet. Res. 33: 1165 (1972).
9. Hsu, F. S., et al. Lead inclusion bodies in osteoclasts. Science 181: 447 (1973).
10. Goyer, R. A., and Rhyne, B. Pathological effects of lead. Internatl. Rev. Exp. Pathol. 12: 1 (1973).
11. Sandstead, H. H., Michelakis, A. M., and Temple, T. E. Lead intoxication: its effect on renin-aldosterone response to sodium deprivation. Arch. Environ. Health 20: 356 (1970).
12. Sobel, A. E., et al. The biochemical behavior of lead. I. Influence of calcium, phosphorus and vitamin D on lead in blood and bone. J. Biol. Chem. 132: 239 (1940).
13. Rosen, J. F., and Roginsky, M. Lead intoxicated children: Plasma levels of 25-hydroxycholecalciferol (25-HCC). Pediatr. Res. 7: 393 (1973).
14. Omdahl, J. L., and DeLuca, H. F. Rachitogenic activity of dietary strontium. I. Inhibition of intestinal calcium absorption and 1,25-dihydroxycholecalciferol synthesis. J. Biol. Chem. 247: 5520 (1972).
15. Diez-Edwald, M., Weintraub, L. R., and Crosby, W. H., 1968. Interrelationship of iron and man-

ganese metabolism. Proc. Soc. Exp. Biol. Med. 129: 448 (1968).
16. Pollack, S., et al. The absorption of nonferrous metals in iron deficiency. J. Clin. Invest. 44: 1470 (1965).
17. Valberg, L. S., Ludwig, J., and Olatunbosun, D. Alteration in cobalt absorption in patients with disorders of iron metabolism. Gastroenterology 56: 241 (1969).
18. Waxman, H. S. and Rabinovitz, M. Control of reticulocyte polyribosome content and hemoglobin synthesis by heme. Biochim. Biophys. Acta 129: 369 (1966).
19. National Academy of Science—National Research Council. Recommended Daily Allowances. 7th Ed., Publication 1694. Washington, D. C., 1968.
20. Ten-State Nutrition Survey, 1968–1970. V. Dietary. U. S. Dept. HEW, Atlanta, Ga., 1972.
21. Skidmore, K. Study of Head Start pre-school children's food habits, Baltimore, Md., (1965); cited in J. L. Kelsay. A compendium of nutritional status studies and dietary evaluation studies conducted in the United States, 1957–1967. J. Nutr. 99: 119 (1969).
22. Stubbs, A. Food use and potential nutritional level of 1225 Texas families. Texas A. & M. University Experiment Station Bulletin No. H-B-1033, 1965.
23. Kolbye, A. Food exposures to lead and their implications. Environ. Health Perspect. No. 7: 65 (1974).
24. Mitchell, D. Lead content of foodstuffs. Environ. Health Perspect. No. 7: 59 (1974).